Modified mTEC Agar, Colilert®, and M-FC Agar - Field Trial Comparison of Bacteria Enumeration Methods in Surface Waters of Eastern Wyoming

Eric Hargett and Lanny Goyn
Wyoming Department of Environmental Quality - Water Quality Division

Introduction

The main criterion for assessment of the potential public health risk of recreational waters in Wyoming is the density of fecal-indicator bacteria in the water column. Currently, the Wyoming Department of Environmental Quality, Water Quality Division (WDEQ/WQD) uses fecal-coliform bacteria as the indicator organisms for assessment of whether recreational uses are maintained in Wyoming's ambient waters (WDEQ/WQD, 2001). WDEQ/WQD proposes to change the indicator organism from fecal-coliform bacteria to *Escherichia coli* (*E. coli*) during the next triennial review of the State of Wyoming's water quality standards. Recognizing that implementation of the revised recreational standard will require adoption of an appropriate method(s) to determine *E. coli* densities, WDEQ/WQD initiated field trials in 2003 to evaluate the performance of two widely recognized and accepted methods for the enumeration of *E. coli* in ambient and effluent waters of eastern Wyoming. The two procedures: Modified mTEC agar and IDEXX's Colilert®, have been adopted as standard methods for monitoring recreational water quality by the United States Environmental Protection Agency (USEPA, 2003).

The Modified mTEC agar procedure is a membrane filtration (MF) method, which produces quantifiable results in 24 hours, provides a direct enumeration of *E. coli* densities, and has been adopted by other state and federal agencies as the enumeration method of choice for *E. coli.* The other method, Colilert®, is a relatively recent most probable number (MPN) procedure that produces results in 24 hours and provides enumerations of *E. coli* densities via interpolation from MPN probability tables. Though results are interpolated, studies have shown that the Colilert® produces results equivalent to those derived from most standard MF methods (Budnick et al., 2001; Cowburn et al., 1994; Eckner, 1998; and Edberg et al., 1989).

Specific objectives of this study were to: 1) address whether results derived from the Colilert® and Modified mTEC methods are comparable for the enumeration of *E. coli* over a broad range of densities in waters of eastern Wyoming; and, 2) compare results from the Colilert® and Modified mTEC methods to those of the M-FC fecal-coliform bacteria method currently used by the WDEQ/WQD.

Methods

Waterbodies in eastern Wyoming that were part of the pre-planned WDEQ/WQD 2003 monitoring schedule for *E. coli* and fecal-coliform bacteria were used as sample sites for this study. Colilert® (N=93), Modified mTEC (N=64), and M-FC (N=94) samples were collected from 28 sites between May and October 2003 (Table 1). Twenty-three of these sites were sampled in ambient recreational waters while the remaining five sites were effluent samples collected from municipal wastewater treatment facilities (Table 1). Though the primary focus of the study was focused on ambient waters, the inclusion of effluent samples from municipal wastewater treatment facilities allowed for the evaluation of fecal-indicator bacteria over a wider range of densities.

Samples were collected with the use of 125 milliliter (mL) Whirl-Pak® bags using standard bacteria collection methods (WDEQ/WQD, 2001a). To obtain sufficient sample volumes for this study, samples were collected in multiple Whirl-Pak® bags at each station for each sample collection period. All samples were preserved on ice and processed at the WDEQ/WQD Water Quality Laboratory in Cheyenne, WY within 6 hours of collection (WDEQ/WQD, 2001a).

Processing and incubation of plated Modified mTEC agar plates followed procedures recommended by USEPA (2000). Processing and incubation of plated M-FC agar plates followed standard WDEQ/WQD procedures (WDEQ/WQD, 2001a). Bacterial colonies on Modified mTEC and M-FC agar plates were enumerated according to methods adopted by WDEQ/WQD (2001a). Processing, incubation, and enumeration of samples for the Colilert® method followed recommended procedures as described by the manufacturer IDEXX (2001). Results from all methods are reported as colonies per 100 mL (col/100 mL).

Non-parametric statistical methods were used to test for correlations and statistical differences between method types. Non-parametric tests were used because fecal-indicator bacteria data were not normally distributed. Wilcoxon's paired rank sample test was used to determine whether results between methods were significantly different. Spearman's rank correlation procedure was used to measure the association and magnitude of the relation between methods. In all tests, a P-value of 0.05 was employed and all analyses were conducted using STATISTICA Version 6 (Statsoft, 2001).

Results

Escherichia coli densities among all sites, as determined from both Colilert® and Modified mTEC methods, ranged from 1 to 35,000 col/100 mL (Table 2). Fecal-coliform densities among all sites, as determined by

the M-FC method, ranged from 1 to 56,000 col/100 mL. The maximum densities for both *E. coli* and fecal-coliform bacteria were primarily found in samples collected from municipal wastewater effluents and urban stream reaches.

The median *E. coli* density among samples where both Colilert® and Modified mTEC methods were applied was 19 col/100 mL for each method. Among samples where Colilert® and M-FC methods were applied, the median density for *E. coli* and fecal-coliform bacteria were 26 and 19 col/100 mL, respectively. Similarly, among samples where both Modified mTEC and M-FC methods were applied, the median density for *E. coli* and fecal-coliform bacteria were 29 and 39 col/100 mL, respectively.

Spearman rank correlation coefficients were significant ($P<0.05$) among all methods. Correlation coefficients between methods were 0.956 for Modified mTEC and M-FC (Figure 1), 0.923 for Colilert® and M-FC (Figure 2), and 0.952 for Colilert® and Modified mTEC (Figure 3).

The Wilcoxon paired rank sample test indicated no significant difference ($P = 0.579$) in *E. coli* enumerations between Colilert® and Modified mTEC methods. Comparison of M-FC fecal-coliform and Colilert® *E. coli* enumerations were not significant ($P = 0.503$), though Colilert® did provide slightly higher median enumerations of *E. coli* relative to fecal-coliform bacteria. The comparison in fecal-indicator enumerations between the Modified mTEC and M-FC methods was found to be significant ($P = 0.005$). The significant difference was due to greater enumerations of fecal-coliform relative to *E. coli.*

Discussion

Comparison of M-FC enumerations to those of Colilert® and Modified mTEC

Spearman's rank correlation revealed strong significant correlations between fecal-coliform and *E. coli* methods. One would expect significant, positive correlations between the M-FC and two *E. coli* enumeration methods due to the fact that *E. coli* is a subset of fecal-coliform. However, strong correlations between fecal-coliform and *E. coli* densities derived from pooled datasets do not necessarily ensure that the relationship will be the same for individual streams, due in part to differences in sources of fecal-indicator bacteria (Clark and Gamper, 2003).

The Wilcoxon paired rank sample test produced somewhat conflicting results when bacteria densities between the two *E. coli* enumeration tests and the M-FC procedure were compared. This study found that in the comparison of M-FC and Modified mTEC methods, fecal-coliform densities were significantly greater than those of *E. coli.* However, densities of *E. coli* were not significantly different compared to fecal-

coliform in the comparison of M-FC and Colilert® methods.

Though it was assumed the comparison between the Modified mTEC and M-FC methods would yield a similar result, the significantly greater fecal-coliform to *E. coli* densities between the aforementioned two methods may be due to several factors.

Initially it was thought that the number of paired samples used in each method comparison could account for the differences in significance between the two method comparisons. Specifically, 84 paired samples were used in the comparison of the Colilert® and M-FC methods, versus 57 paired samples used in the Modified mTEC and M-FC comparison. Of the samples used in each comparison, 47 were common to both. Wilcoxon paired rank sample tests performed on the Modified mTEC/M-FC and Colilert®/M-FC comparisons using only the 47 paired samples common among all methods yielded similar results. Another reason for the differences may be the use of bacteria enumerations based on extrapolations from fractional plate counts. Some samples with appreciably high numbers of fecal-coliform bacteria were enumerated based on ½ or ¼ plate counts. In other words, the plated bacteria colonies were so numerous, that identification of all individual colonies for the entire agar plate was difficult. Therefore, the ½ or ¼ section of the plate where individual colonies were the easiest to identify were enumerated. Counts from these plate sections were then multiplied by the appropriate multiplier (i.e., 2 for the ½ plate count, 4 for the ¼ plate count) to arrive at the final enumeration for the entire plate. The distribution of bacteria colonies across the agar plate is usually non-uniform, thus total bacteria counts may vary considerably for an individual plate depending on what ½ or ¼ section of the plate is chosen for the enumeration. In hindsight, the use of smaller sample volumes or dilutions would have eliminated the need for fractional plate counts and may have resulted in more similar results between the Modified mTEC and M-FC methods.

The significant difference in enumerations between the Modified mTEC and M-FC methods may also be due to reasons inherently associated with MF methods. According to Budnick et al., (2001) as well as experience within WDEQ/WQD, inaccurate counts from MF methods could result from the individual or combined effects of these and other factors: variations in filter quality; filtering of highly turbid samples (which characterized several samples used in this study) can concentrate inorganic and organic particulate matter on the filter surface making identification of colonies difficult; and/or insufficient rinsing of the aliquot sample chambers during filtration. Eaton et al., (1998) also states that low bacteria enumerations obtained from MF methods may be caused by the presence of high numbers of non-coliforms or toxic substances in the sample. Interference from non-coliforms do not appear to influence Colilert® results since the media contains two nutrient-indicators that are metabolized by coliform enzymes, which are absent in most non-coliform organisms (IDEXX, 2001). The few non-coliforms that do produce these enzymes are selectively

suppressed by Colilert's® specifically formulated media (IDEXX, 2001).

Comparison of Modified mTEC and Colilert®

Analysis of bacteria data indicated that no significant difference existed in the *E. coli* enumeration results between the Modified mTEC and Colilert® methods. In addition, mean and median bacteria densities between the two results were essentially equal. Both the Modified mTEC and Colilert® methods provided statistically equivalent performances in this field trial study. Furthermore, correlation coefficients show a strong significant relationship between Modified mTEC and Colilert®. These results suggest that either method could confidently be used to enumerate *E. coli* densities in treated sewage and ambient surface waters.

Although both *E. coli* enumeration methods tested in this study were found to provide equivalent and accurate enumerations when a sufficient number of aliquots per sample were processed, the Colilert® method had several advantages in processing and equipment requirements compared to the Modified mTEC method. One of the greatest advantages of Colilert® is that samples are processed in a fraction of the time it takes to process samples using the Modified mTEC method. Secondly, although the Colilert® trays require significantly more incubator space compared to the Modified mTEC plates, the single incubation temperature eliminates the need for two incubators required for the Modified mTEC method. Third, many of the processing steps and limitations of MF methods described previously are removed and/or do not influence final results with the Colilert® method. Many of these advantage comparisons support observations made by Budnick et al., (2001), Ostensvik (2000), and Eckner (1998).

Literature Cited

Budnick, G. E., R. T. Howard, and D. Mayo. 2001. Comparison of Colilert-18 to the mTEC agar method for the enumeration of *Escherchia coli* in recreational waters. Proceedings from the American Society of Microbiologists Annual Convention May 2001.

Clark, M.L. and M.E. Gamper. 2003. A synoptic study of fecal-indicator bacteria in the Wind River, Bighorn River, and Goose Creek Basins, Wyoming, June-July 2000. U.S. Geological Survey. Water-Resources Investigations Report 03-4055.

Cowburn, J.K., T. Goodall, E.J. Fricker, K.S. Walter, and C.R. Fricker. 1994. A preliminary study of the use of Colilert for water quality monitoring. Applied Microbiology 19:50-52.

Eaton, A.D., L.S. Cleseri, and A.E. Greenberg. 1998. Standard Methods for the Examination of Water and Wastewater. American Public Health Association, Washington, D.C.

Eckner, K.F. 1998. Comparison of membrane filtration and multiple-tube fermentation by the Colilert and Enterolert methods for detection of waterborne coliform bacteria, *Escherchia coli*, and enterococci used in drinking and bathing water quality monitoring in southern Sweden. Applied and Environmental Microbiology 64:3079-3083.

Edberg, S.C., M.J. Allen, D.B. Smith, and the National Collaborative Study. 1989. National field evaluation of a defined substrate method for the simultaneous detection of total coliforms and *Eshcherichia coli* form drinking water: comparison with presence-absence techniques. Applied Environmental Microbiology 55:1003-1008.

Fricker, E. J. and C.R. Fricker. 1996. Use of two presence/absence systems for the detection of *E. coli* and coliforms from water. Water Resources 30:2226-2228.

IDEXX. 2001. Colilert Test Kit Procedure. IDEXX Laboratories, Inc., One IDEXX Drive, Westbrook, Maine 04092. www.idexx.com

Ostensvik, O. 2000. Coliform bacteria and *Escherchia coli* in Norwegian drinking water sources-comparison of methods based on the fermentation of lactose and methods based on the activity of specific enzymes. AWWA Water Quality Technology Conference Proceedings, Salt Lake City, Utah.

Statsoft. 2001. STATISTICA System Reference. Statsoft, Inc., 2300 East 14th Street, Tulsa, Oklahoma, 74104.

USEPA. 2000. Improved enumeration methods for the recreational water quality indicators: Enterococci and *Escherichia coli*. Office of Science and Technology, Washington D.C. 20460. EPA/821/R-97/004.

USEPA. 2003. Guidelines Establishing Test Procedures for the Analysis of Pollutants; Analytical Methods for Biological Pollutants in Ambient Waters; Final Rule. Federal Register, Vol. 68, No. 139. FRL-7529-7.

WDEQ/WQD. 2001. Wyoming Water Quality Rules and Regulations, Chapter 1, Wyoming Surface Water Quality Standards. Water Quality Division, Cheyenne, WY.

WDEQ/WQD. 2001a. Manual of Standard Operating Procedures for Sample Collection and Analysis. Wyoming Department of Environmental Quality, Water Quality Division, Watershed Program, Cheyenne, WY.

Table 1. Descriptive information for WDEQ/WQD sample sites, May-October 2003. NOTE: WWTF equates to a waste-water treatment facility.

SITE NAME	LEGAL LOCATION
City of Casper, WY WWTF Outfall 001	NWNW of Sec. 2, T33N, R79W
City of Douglas, WY WWTF Outfall 001	SWNE of Sec. 8, T32N, R71W
City of Laramie, WY WWTF Outfall 001	NWNW of Sec. 16, T16N, R73W
City of Wheatland, WY WWTF Outfall 001	SWNE of Sec. 1, T24N, R68W
Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	NWNW of Sec. 6, T13N, R66W
Crow Creek at Morrie Avenue, City of Cheyenne, WY	NWSE of Sec. 5, T13N, R66W
Dry Creek at College Drive, City of Cheyenne, WY	NWSW of Sec. 26, T14N, R66W
Dry Creek at Dell Range Boulevard, City of Cheyenne, WY	NWNW of Sec. 29, T14N, R66W
Laramie River above City of Laramie, WY WWTF Outfall 001	NWNW of Sec. 16, T16N, R73W
Laramie River below City of Laramie, WY WWTF Outfall 001	NWNW of Sec. 16, T16N, R73W
Medicine Bow River above City of Medicine Bow, WY WWTF Outfall 001	SENW of Sec. 5, T22N, R78W
Middle Crow Creek above US Forest Service Road 700	SESW of Sec. 14, T14N, R71W
Minnehaha Lake at Holiday Park in Cheyenne, WY	NESW of Sec. 32, T14N ,R66W
N. Branch Crow Creek above Upper North Crow Reservoir	SENE of Sec. 25, T15N, R71W
N. Branch Crow Creek above WY Highway 210	NENE of Sec. 32, T15N, R71W
N. Branch Crow Creek below US Forest Service Road 701	NWNW of Sec. 34, T15N, R71W
North Platte River above City of Casper, WY WWTF Outfall 001	NWNW of Sec. 2, T33N, R79W
North Platte River above City of Douglas, WY WWTF Outfall 001	SWNE of Sec. 8, T32N, R71W
North Platte River above Town of Guernsey, WY WWTF Outfall 001	NWSE Sec. 2, T27N, R66W
North Platte River below City of Casper, WY WWTF Outfall 001	NWNW of Sec. 2, T33N, R79W
North Platte River below City of Douglas, WY WWTF Outfall 001	SWNE of Sec. 8, T32N, R71W
Rawhide Creek above Town of Lingle, WY WWTF Outfall 001	NWNE of Sec. 29, T25N, R62W
Rawhide Creek below Town of Lingle, WY WWTF Outfall 001	NWNE of Sec. 29, T25N, R62W
S. Branch Crow Creek above Upper North Crow Reservoir	SENE of Sec. 36, T15N, R71W
S. Branch Crow Creek below WY Highway 210	NWNE of Sec. 3, T14N, R71W
Town of Lingle, WY WWTF Outfall 001	NWNE of Sec. 29, T25N, R62W
Wheatland Creek above City of Wheatland, WY WWTF Outfall 001	SWNE of Sec. 1, T24N, R68W
Wheatland Creek below City of Wheatland, WY WWTF Outfall 001	SWNE of Sec. 1, T24N, R68W

Table 2. Fecal-indicator bacteria results for WDEQ/WQD sample sites, May-October 2003. NOTE: col/100 mL equates to colonies per 100 milliliters and WWTF equates to a waste-water treatment facility.

DATE SAMPLED	TIME SAMPLED	SITE NAME	Fecal Coliform m-FC media col/100 mL	*E. coli* mTEC agar col/100 mL	*E. coli* Colilert Media col/100 mL
5/13/2003	1225	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	10	14	15
5/13/2003	1240	Crow Creek at Morrie Avenue, City of Cheyenne, WY	2	12	39
5/14/2003	1155	Middle Crow Creek above US Forest Service Road 700	6	5	3
5/14/2003	1020	N. Branch Crow Creek above Upper North Crow Reservoir	2	1	3
5/14/2003	0845	N. Branch Crow Creek above WY Highway 210	1	2	2
5/14/2003	0925	N. Branch Crow Creek below US Forest Service Road 701	2	1	1
5/14/2003	1125	S. Branch Crow Creek above Upper North Crow Reservoir	1	1	1
5/14/2003	0935	S. Branch Crow Creek below WY Highway 210	1	1	1
5/20/2003	1152	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	49	60	56
5/20/2003	1205	Crow Creek at Morrie Avenue, City of Cheyenne, WY	228	80	93
5/20/2003	1110	Middle Crow Creek above US Forest Service Road 700	3	2	4
5/20/2003	0953	N. Branch Crow Creek above Upper North Crow Reservoir	5	2	6
5/20/2003	0845	N. Branch Crow Creek above WY Highway 210	6	5	9
5/20/2003	0905	N. Branch Crow Creek below US Forest Service Road 701	17	11	12
5/20/2003	0905	N. Branch Crow Creek below US Forest Service Road 701	14	8	NM
5/20/2003	1042	S. Branch Crow Creek above Upper North Crow Reservoir	5	1	1
5/20/2003	0923	S. Branch Crow Creek below WY Highway 210	2	1	1
5/22/2003	0940	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	35	42	45
5/22/2003	0925	Crow Creek at Morrie Avenue, City of Cheyenne, WY	153	140	82
5/27/2003	1205	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	60	70	179
5/27/2003	1155	Crow Creek at Morrie Avenue, City of Cheyenne, WY	130	63	57
5/27/2003	1100	Middle Crow Creek above US Forest Service Road 700	11	13	12
5/27/2003	1027	N. Branch Crow Creek above Upper North Crow Reservoir	10	6	5
5/27/2003	0845	N. Branch Crow Creek above WY Highway 210	15	21	15
5/27/2003	0900	N. Branch Crow Creek below US Forest Service Road 701	14	21	12
5/27/2003	0900	N. Branch Crow Creek below US Forest Service Road 701	14	6	11
5/27/2003	0945	S. Branch Crow Creek above Upper North Crow Reservoir	7	3	14
5/27/2003	0915	S. Branch Crow Creek below WY Highway 210	5	10	3
6/3/2003	1225	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	115	74	118
6/3/2003	1215	Crow Creek at Morrie Avenue, City of Cheyenne, WY	230	150	147
6/3/2003	1100	Middle Crow Creek above US Forest Service Road 700	8	8	12
6/3/2003	1027	N. Branch Crow Creek above Upper North Crow Reservoir	11	6	10
6/3/2003	0845	N. Branch Crow Creek above WY Highway 210	65	14	89
6/3/2003	0905	N. Branch Crow Creek below US Forest Service Road 701	23	21	32
6/3/2003	0945	S. Branch Crow Creek above Upper North Crow Reservoir	7	15	21
6/3/2003	0945	S. Branch Crow Creek above Upper North Crow Reservoir	16	22	14
6/3/2003	0915	S. Branch Crow Creek below WY Highway 210	12	13	11
6/5/2003	0920	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	NM	700	649
6/5/2003	0910	Crow Creek at Morrie Avenue, City of Cheyenne, WY	NM	1300	649
6/5/2003	0855	Dry Creek at College Drive, City of Cheyenne, WY	NM	3000	2420
6/5/2003	0835	Minniehaha Lake at Holiday Park in Cheyenne, WY	NM	18	16
6/10/2003	1050	Middle Crow Creek above US Forest Service Road 700	9	NM	12
6/10/2003	1015	N. Branch Crow Creek above Upper North Crow Reservoir	7	1	11
6/10/2003	0830	N. Branch Crow Creek above WY Highway 210	42	22	32
6/10/2003	0850	N. Branch Crow Creek below US Forest Service Road 701	33	20	32
6/10/2003	0935	S. Branch Crow Creek above Upper North Crow Reservoir	26	NM	19
6/10/2003	0905	S. Branch Crow Creek below WY Highway 210	10	NM	9
6/12/2003	1000	Crow Creek at Martin Luther King Jr. Park, City of Cheyenne, WY	NM	163	361
6/12/2003	0950	Crow Creek at Morrie Avenue, City of Cheyenne, WY	NM	264	276
6/12/2003	0935	Dry Creek at College Drive, City of Cheyenne, WY	1200	610	980
6/12/2003	0925	Dry Creek at Dell Range Boulevard, City of Cheyenne, WY	NM	107	91
6/17/2003	1040	City of Wheatland, WY WWTF Outfall 001	240	170	NM
6/17/2003	1050	Wheatland Creek above City of Wheatland, WY WWTF Outfall 001	7637	3091	NM
6/17/2003	1130	Wheatland Creek below City of Wheatland, WY WWTF Outfall 001	43637	2017	NM
6/18/2003	1110	North Platte River above Town of Guernsey, WY WWTF Outfall 001	42	47	NM

NM- Not measured

Table 2 (cont.). Fecal-indicator bacteria results for WDEQ/WQD sample sites, May-October 2003. NOTE: col/100 mL equates to colonies per 100 milliliters and WWTF equates to a waste-water treatment facility.

DATE SAMPLED	TIME SAMPLED	SITENAME	Fecal Coliform m-FC media col/100 mL	*E. coli* mTEC agar col/100 mL	*E. coli* Colilert Media col/100 mL
7/8/2003	0950	City of Laramie, WY WWTF Outfall001	40	40	NM
7/8/2003	1050	Laramie River above City of Laramie, WY WWTF Outfall 001	207	143	NM
7/8/2003	1105	Laramie River below City of Laramie, WY WWTF Outfall 001	200	130	NM
7/9/2003	1045	Rawhide Creek above Town of Lingle, WY WWTF Outfall 001	370	460	435
7/9/2003	1055	Rawhide Creek below Town of Lingle, WY WWTF Outfall 001	665	440	411
7/9/2003	1030	Town of Lingle, WY WWTF Outfall001	56000	35000	α>2420
7/10/2003	1210	Medicine Bow River above City of Medicine Bow, WY WWTF Outfall001	230	170	118
7/10/2003	1210	Medicine Bow River above City of Medicine Bow, WY WWTF Outfall001	330	160	NM
9/15/2003	1245	Middle Crow Creek above US Forest Service Road 700	140	NM	130
9/15/2003	1205	N. Branch Crow Creek above Upper North Crow Reservoir	90	115	114
9/15/2003	1005	N. Branch Crow Creek above WY Highway 210	85	100	133
9/15/2003	1030	N. Branch Crow Creek below US Forest Service Road 701	4400	2740	2420
9/15/2003	1120	S. Branch Crow Creek above Upper North Crow Reservoir	4	NM	3
9/15/2003	1050	S. Branch Crow Creek below WY Highway 210	7	4	11
9/18/2003	1125	Middle Crow Creek above US Forest Service Road 700	420	NM	613
9/18/2003	1050	N. Branch Crow Creek above Upper North Crow Reservoir	80	NM	86
9/18/2003	0915	N. Branch Crow Creek above WY Highway 210	100	NM	96
9/18/2003	0930	N. Branch Crow Creek below US Forest Service Road 701	3300	NM	4840
9/18/2003	1010	S. Branch Crow Creek above Upper North Crow Reservoir	14	NM	11
9/18/2003	0945	S. Branch Crow Creek below WY Highway 210	28	NM	32
9/23/2003	1100	Middle Crow Creek above US Forest Service Road 700	106	NM	96
9/23/2003	1100	Middle Crow Creek above US Forest Service Road 700	101	NM	116
9/23/2003	1030	N. Branch Crow Creek above Upper North Crow Reservoir	20	NM	44
9/23/2003	0905	N. Branch Crow Creek above WY Highway 210	36	NM	66
9/23/2003	0915	N. Branch Crow Creek below US Forest Service Road 701	767	NM	3030
9/23/2003	0950	S. Branch Crow Creek above Upper North Crow Reservoir	0	NM	2
9/23/2003	0925	S. Branch Crow Creek below WY Highway 210	14	NM	16
9/25/2003	1200	Middle Crow Creek above US Forest Service Road 700	290	NM	313
9/25/2003	1125	N. Branch Crow Creek above Upper North Crow Reservoir	55	NM	77
9/25/2003	1010	N. Branch Crow Creek above WY Highway 210	420	NM	31
9/25/2003	1023	N. Branch Crow Creek below US Forest Service Road 701	400	NM	579
9/25/2003	1047	S. Branch Crow Creek above Upper North Crow Reservoir	9	NM	1
9/25/2003	1030	S. Branch Crow Creek below WY Highway 210	8	NM	11
9/29/2003	1105	Middle Crow Creek above US Forest Service Road 700	253	NM	214
9/29/2003	1030	N. Branch Crow Creek above Upper North Crow Reservoir	195	NM	248
9/29/2003	0910	N. Branch Crow Creek above WY Highway 210	70	NM	117
9/29/2003	0920	N. Branch Crow Creek below US Forest Service Road 701	720	NM	579
9/29/2003	0950	S. Branch Crow Creek above Upper North Crow Reservoir	4	NM	1
9/29/2003	0927	S. Branch Crow Creek below WY Highway 210	12	NM	9
10/1/2003	1315	City of Douglas, WY WWTF Outfall001	250	NM	1733
10/1/2003	1125	City of Casper, WY WWTF Outfall 001	32500	NM	α>2420
10/1/2003	1120	North Platte River above City of Casper, WY WWTF Outfall001	37	NM	38
10/1/2003	1310	North Platte River above City of Douglas, WY WWTF Outfall001	7	NM	11
10/1/2003	1130	North Platte River below City of Casper, WY WWTF Outfall001	1934	NM	1203
10/1/2003	1130	North Platte River below City of Casper, WY WWTF Outfall001	NM	NM	1844
10/1/2003	1305	North Platte River below City of Douglas, WY WWTF Outfall001	17	NM	5
10/1/2003	1305	North Platte River below City of Douglas, WY WWTF Outfall001	3	NM	10
NM- Not measured					
α - Value not used in comparative analysis.					

Figure 1. Modified mTEC and M-FC correlation plot, May-October 2003

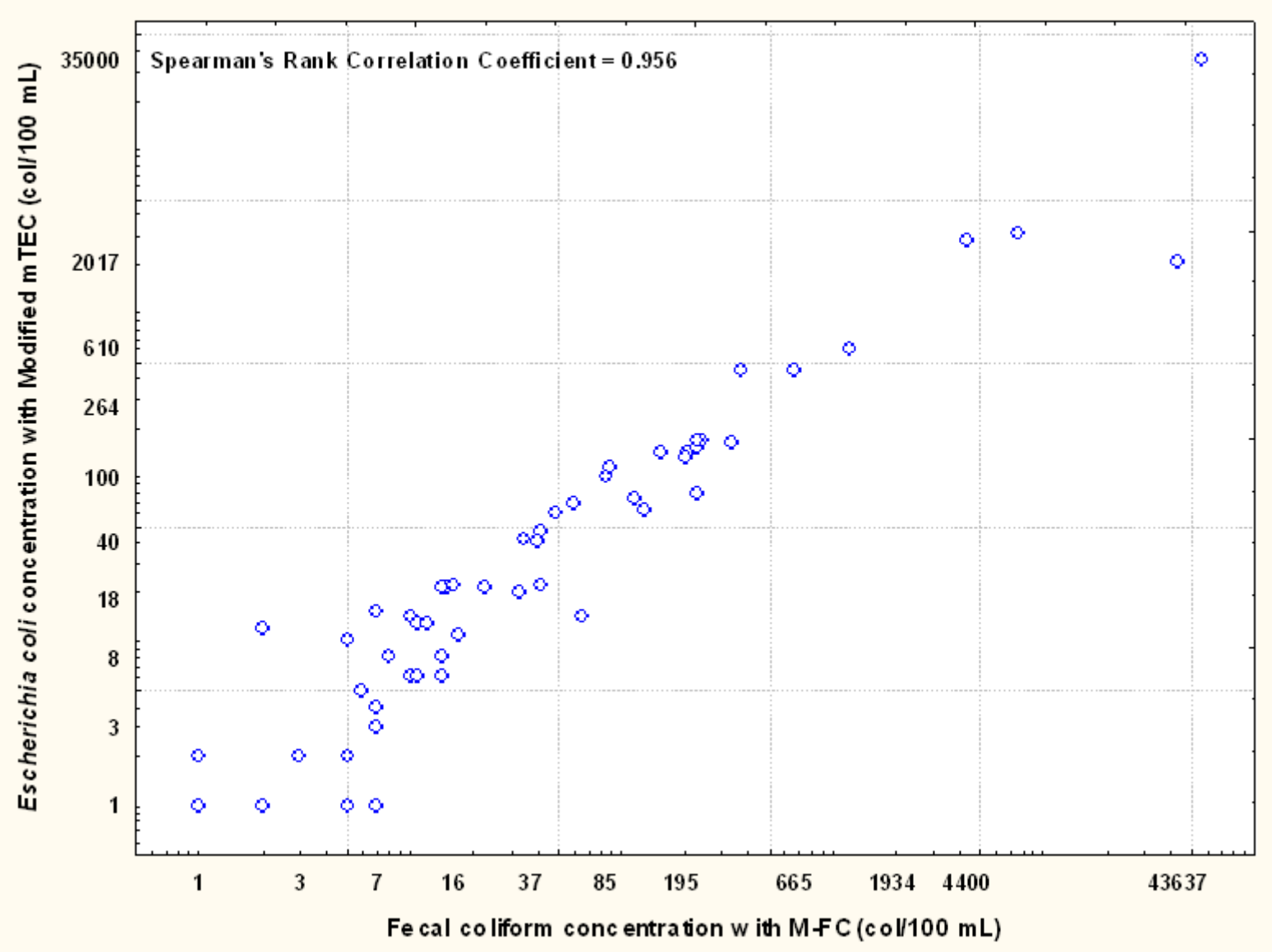

Figure 2. Colilert® and M-FC correlation plot, May-October 2003.

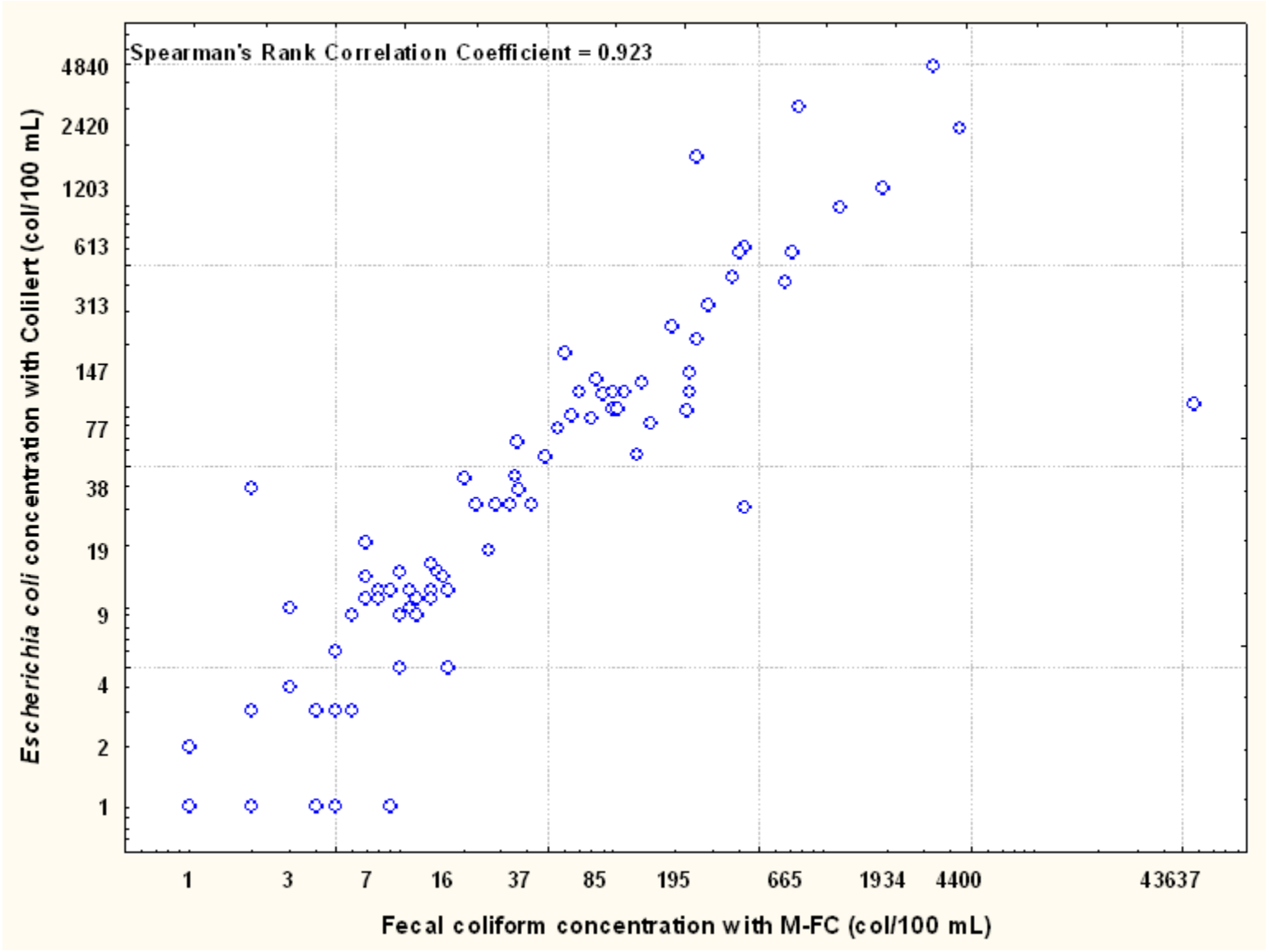

Figure 3. Colilert® and Modified mTEC correlation plot, May-October 2003.